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THE RESPONSES OF AORTIC ARCH AND RIGHT SUBCLAVIAN BARORECEPTORS TO CHANGES OF NON-PULSATILE PRESSURE AND THEIR MODIFICATION BY HYPOTHERMIA

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SUMMARY

1. A method is described for isolation of the aortic arch and right subclavian–carotid angle *in situ* in the rabbit and perfusion with Krebs–Henseleit solution or blood under controlled conditions of pressure and temperature.

2. The characteristics of the baroreceptors of the aortic arch and right subclavian–carotid angle were studied by recording from single or few-fibre preparations of the left and right aortic nerves respectively. Curves were plotted to show the relationship between the frequency of baroreceptor impulse activity and intra-aortic pressure during non-pulsatile perfusion under steady-state conditions.

3. The aortic arch and right subclavian–carotid angle baroreceptors were found to have similar characteristics. Three types of response of the baroreceptors at the threshold pressure to a steady intra-aortic pressure are described.

4. Increasing the intra-aortic pressure increased the frequency of impulses in fibres previously active and caused recruitment of other fibres in multi-fibre preparations. The relationship was linear at low pressures and a point of inflexion occurred at higher pressures in the majority of fibres.

5. Lowering the temperature of the perfusate reduced the impulse frequency at any given pressure.

6. The curves obtained during stepwise increases and decreases in intra-aortic arch pressure were dissimilar, particularly at the lower end of the pressure range. This phenomenon is probably due to properties of the arterial wall.

7. When the aortic arch preparation was excised, changes occurred in

the shape of the impulse frequency–pressure curves from baroreceptors in both areas. The point of inflexion was elevated and a higher percentage of fibres failed to reach a point of inflexion in the pressure range studied.

INTRODUCTION

Our knowledge of the physiological characteristics of arterial baroreceptors stems mainly from studies carried out on the carotid sinuses (Bronk & Stella, 1932, 1935; Landgren, 1952*a*, *b*; see also Heymans & Neil, 1958) and it has been generally assumed that the baroreceptors in the aortic arch show similar characteristics. However, in a recent study it has been shown that whereas the reflex vasomotor responses elicited by changes in mean carotid sinus pressure are modified by alterations of pulse pressure, those evoked by the aortic-arch baroreceptors through changes of mean pressure are only weakly affected by modifications in pulse pressure (Angell James & Daly, 1970). But apart from a few studies of the impulse discharge in baroreceptor fibres from the aortic arch in the entire animal (Adrian, 1926; Bloor, 1964; Aars, 1968), there has been no systematic investigation of their responses to alteration of mean pressure, pulse pressure and pulse frequency under conditions of controlled perfusion.

This paper describes a technique for isolating and perfusing the aortic arch and right subclavian–carotid angle for experimental investigation of baroreceptor nerve fibre activity from these two areas under controlled conditions. The pattern of firing of single and few-fibre recordings of the aortic nerves was studied during perfusion at non-pulsatile pressures. Some of the results of this study have been described briefly elsewhere (Angell James, 1968) and in detail in a thesis (Angell James, 1969).

METHODS

New Zealand white rabbits weighing 1·6–3·5 kg were anaesthetized with 25 % urethane (1·5–2·0 g/kg; British Drug Houses, Ltd.) given intravenously. During this period it was found that music was helpful in keeping the animals quiet and relaxed.

A tracheostomy was performed and the common carotid arteries were prepared for ligation and cannulation.

Isolation of the aortic arch and subclavian regions. Positive pressure ventilation was maintained with a bellows respirator pump (C. F. Palmer, Ltd.) at a frequency of 30 c/min. A mid line thorocotomy was performed and the right and left subclavian arteries and the descending thoracic aorta, below the level of the left pulmonary artery, were prepared for subsequent ligation. The internal mammary arteries were ligated together with any anomalous vessels arising from the aortic arch and its major branches (Angell James, 1969) (Fig. 1). These surgical procedures were facilitated by use of specially designed aneurysm needles (Daly & Angell James, 1968).

Perfusion of the preparation. The right common carotid artery was cannulated with a nylon cannula ('Portex', o.d. 1·65 mm, i.d. 1·19 mm) and heparin was administered intra-arterially ('Pularin', Evans Medical Ltd., 1000 i.u./kg). The right and left subclavian arteries were then ligated whilst recording from the right and left aortic nerves to determine whether this procedure had any effect on impulse activity, as the ligatures were very close to the nerves to the baroreceptor areas.

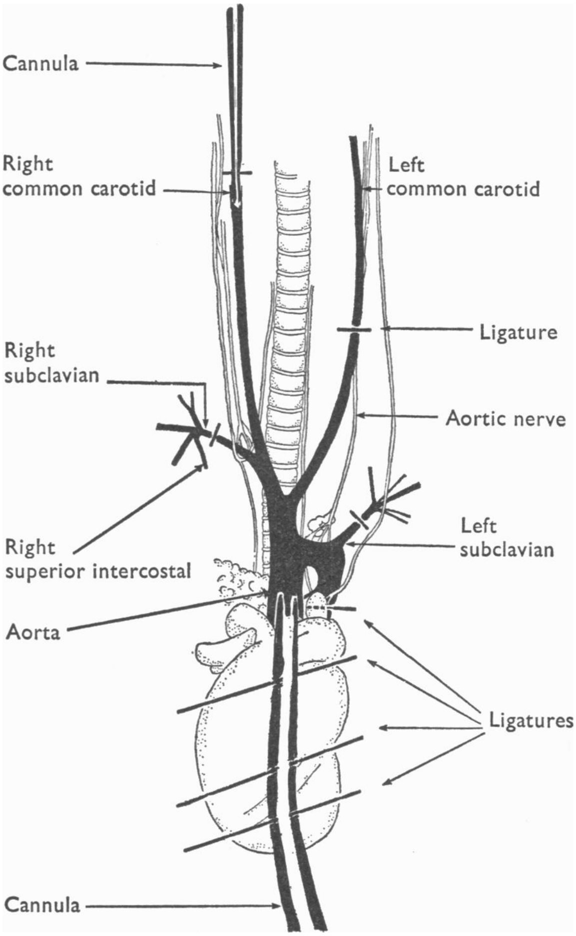

Fig. 1. Diagram showing the isolated perfused aortic arch and right subclavian regions of the rabbit, and indicating the site of the ligatures and cannulae. The architecture of the main branches of the aortic arch is variable (Angell James, 1969), but that depicted here is the commonest variant.

The preparation was perfused through a brass cannula (o.d. 4 mm, i.d. 3·8 mm, length 75 mm), bent in the middle through 40°, which was passed through the wall of the left ventricle so that its tip lay in the ascending aorta. It was secured in position by tapes tied in the atrio-ventricular groove and around the ventricles. The left common carotid artery and the descending thoracic aorta were ligated. The effluent passed out via the right common carotid cannula and the flow was controlled by a screw-clamp-type resistance.

The perfusion fluid was a modified Krebs–Henseleit solution (KCl, 0·42 g; $CaCl_2$ 0·141 g; $MgSO_4.7H_2O$, 0·294 g; $NaH_2PO_4.2H_2O$, 0·183 g; NaCl, 6·87 g; $NaHCO_3$, 2·1 g; glucose, 1 g; de-ionized water to 1 l.). The fluid was equilibrated with 95 % oxygen and 5 % carbon dioxide, giving a pH of 7·4. The flow through the preparation varied between 3 and 15 ml./min.

In some experiments rabbit's blood was used and this contained heparin in a concentration of 20,000 i.u./l. and penicillin, 5000 u./l. In other experiments dog's blood was used containing both heparin and penicillin in the same concentrations.

Temperature control. The perfusion fluid was contained in an aspirator submerged in a thermostatically controlled water-bath at 40° C. This fluid was carried to the preparation in a polyethylene tube, surrounded by a heated water-jacket. The temperature of the perfusate was recorded by means of a copper–constantan thermocouple lying in the tip of the aortic cannula and readings were taken from a galvanometer. The system was calibrated before and after each experiment. The temperature in the aortic arch was maintained constant between 37° and 39° C in different experiments (mean 38·2° C).

In some experiments the effect of lowering the temperature of the perfusate was studied. For this purpose a small cooling coil was placed in the perfusion circuit in parallel to the main perfusion tubing and could therefore be by-passed if necessary.

The mean aortic arch pressure was measured with a mercury manometer via a nylon catheter the tip of which lay in the end of the aortic cannula, and recorded on a smoked-paper kymograph. The pressure was controlled by means of a compressed air–airleak by-pass system on the aspirator.

The 'excised' preparation. In some experiments the preparation perfused *in situ*, as described above, was subsequently excised from the animal and placed in a Perspex box with controlled temperature, as described by Angell James (1971*b*).

Recording of action potentials. Single or few-fibre preparations of the aortic nerves were made and the impulse activity was recorded by means of saline wick silver–silver chloride electrodes connected to a Tektronix 122 preamplifier. The potentials were displayed on one channel of a cathode-ray oscilloscope. The output also passed to a loudspeaker. The second channel of the oscilloscope was used to record a 50-cycle time marker and stimulus marker. Permanent records were obtained by photographing the screen with a Cossor camera using Ilford recording paper (type NS 6).

Analysis of results

Measurement of impulse frequency. Under steady-state conditions the number of impulses recorded in one second was counted. The impulse frequency was then plotted against the measured intra-aortic pressure. It will be seen in Fig. 2 that, during non-pulsatile perfusion, these points fell on a straight line in the lower pressure range, and the 'gradient' or 'slope' of this part of the curve was measured and expressed in impulses/sec.mm Hg. The lowest pressure at which the baroreceptors discharge any impulse is called the 'threshold' pressure.

At a certain pressure the relationship between the impulse frequency and intra-aortic pressure ceased to be linear and this pressure is described as the point of inflexion.

The 'threshold index' was calculated by dividing the frequency of impulses (impulses/sec) at the threshold pressure by the threshold pressure (mm Hg) and expressed as the number of impulses/sec.mm Hg. The index at the point of inflexion was calculated similarly by dividing the frequency of impulses (impulses/sec)

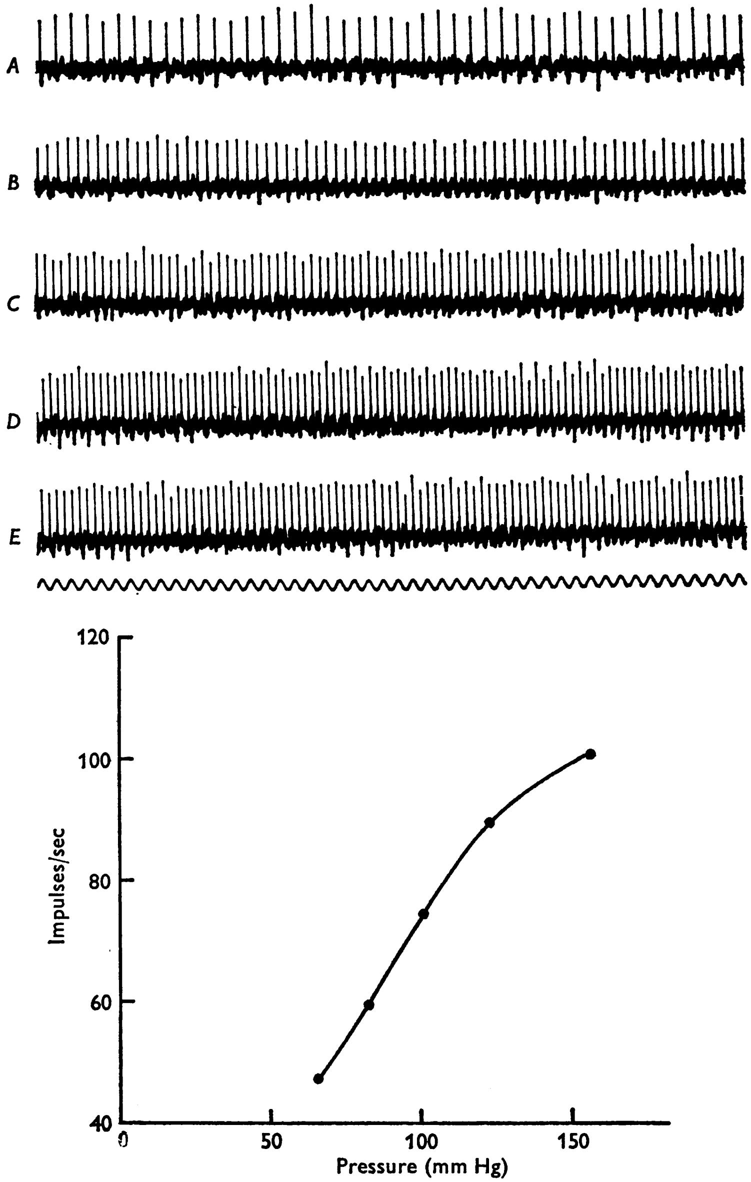

Fig. 2. The relationship between impulse activity in a single baroreceptor fibre of the left aortic nerve and intra-aortic pressure. Rabbit, wt. 1·8 kg. The isolated aortic arch was perfused with Krebs–Henseleit solution at 38·5° C. *A*, impulse frequency 47/sec at its threshold pressure of 65 mm Hg; *B*, impulse frequency 59/sec at a pressure of 82 mm Hg; *C*, impulse frequency 74/sec at a pressure of 100 mm Hg; *D*, impulse frequency 89/sec at a pressure of 122 mm Hg; *E*, impulse frequency of 100/sec at a pressure of 155 mm Hg. Time marker 50 c/s.

by the pressure (mm Hg) at the point of inflexion and expressed as impulses/sec.mm Hg.

Statistical analysis. The results are expressed as mean ± the standard error of the mean (S.E. of mean). The Student *t* test was used to evaluate the significance of the difference of two means of grouped values, and the significance of the difference from zero of the mean of the difference between paired observations.

RESULTS

I. The aortic arch preparation *in situ*

In single- or few-fibre preparations of the aortic nerves it was found that the size of the action potentials was variable. Raising the mean pressure of the perfusate in a stepwise fashion resulted in a sudden increase in impulse activity which was greater than that at which it settled at the new pressure. Although under steady-state conditions considerable variability was found in the shape of the curve relating impulse frequency to intra-aortic arch pressure with different fibres, control tests made on six fibres in six experiments showed that the curves were repeatable for at least 30 min.

Impulse frequency–aortic pressure curves for left and right aortic nerves

Left aortic nerve

The shape of the curve relating impulse frequency to aortic arch pressure differed considerably from one fibre to another but there were certain features which were common to most fibres. When the pressure was raised from zero, no impulses were recorded until a critical pressure was reached, the threshold pressure, which in the experiment depicted in Fig. 2 was 65 mm Hg.

The nature of the discharge in fibres at their threshold pressures was variable and has been classified arbitrarily into three types. In Type 1 there was a critical level of pressure at which the discharge of impulses in the fibre commenced (Fig. 2*A*). In Type 2 the fibres discharged intermittently, in some cases in bursts lasting for a second and then ceasing for a second or two. A third form of discharge (Type 3) was that in which the impulse frequency remained constant over a range of pressure of about 20–40 mm Hg before increasing as the pressure increased. In some of these cases the frequency of the fibre even diminished by 2 or 3 impulses/sec or more before the stage of increasing with the intra-aortic pressure. Of the twenty-six fibres studied, sixteen were of Type 1, four of Type 2 and six of Type 3.

The results of an analysis of twenty-six single- or few-fibre preparations of the left aortic nerve in fifteen experiments are shown in Table 1. The threshold pressure of the fibres varied from 0 to 118 mm Hg (mean

50·8 ± 6·0), and have been plotted as a block diagram in Fig. 4 (left). It is evident that most fibres have a threshold in the range of pressure of 10–70 mm Hg.

TABLE 1. Characteristics of the curves relating impulse activity to intra-aortic pressure from baroreceptor fibres of the left and right aortic nerves with the aortic arch perfused (*A*) *in situ* and (*B*) excised

	(*A*) *In situ* Aortic nerve		(*B*) Excised Aortic nerve	
	Left	Right	Left	Right
No. of experiments	15	5	16	6
No. of fibres	26	10	31	10
Threshold pressure (mm Hg)	50·8 ± 6·0 (0–118)	53·2 ± 9·2 (0–104) $P > 0·8$	52·9 ± 4·5 (15–110)	50·9 ± 7·0 (15–95)
Threshold frequency (impulses/sec)	35·7 ± 2·2 (12–59)	33·0 ± 6·9 (7–71) $P > 0·6$	29·9 ± 2·4 (7–58)	22·3 ± 3·8 (4–39)
Threshold index (impulses/sec.mm Hg)	3·18 ± 2·24 (0·13–59·0)	4·28 ± 3·29 (0·07–34·0) $P > 0·7$	0·79 ± 0·16 (0–2·33)	0·63 ± 0·23 (0·07–2·60)
Gradient (impulses/sec.mm Hg)	1·11 ± 0·14 (0·28–2·85)	1·13 ± 0·14 (0·58–2·2) $P > 0·9$	1·58 ± 0·05 (0·29–3·56)	1·34 ± 0·28 (0·36–2·77)
Point of inflexion (mm Hg)	116·3 ± 6·8 (63–158)	97·7 ± 5·5 (80–121) $P > 0·1$	107·2 ± 7·5 (70–167)	74 (70 and 78)
Frequency at the point of inflexion (impulses/sec)	77·2 ± 6·0 (35–118)	65·5 ± 5·3 (48–95) $P > 0·2$	88·1 ± 9·8 (31–160)	66·5 (63 and 70)
Index at point of inflexion (impulses/sec.mm Hg)	0·68 ± 0·055 (0·22–0·91)	0·7 ± 0·11 (0·4–1·19) $P > 0·7$	0·87 ± 0·10 (0·2–1·58)	0·9 (0·9 and 0·9)
Fibres without point of inflexion				
No.	10	4	16	8
%	38·5	40·0	51·6	80·0

The open values are the means ± S.E. of mean. The ranges are in parentheses.

The first part of the curves relating impulse frequency to intra-aortic pressure was usually linear (Figs. 2, 3) after which increments of pressure produced smaller increases in impulse frequency or even a decrease. In some cases the discharge became intermittent or ceased altogether. This point of inflexion was not obtained for ten fibres either because it did not fall within the recording range of pressure even at pressures in access of

250 mm Hg or because the fibre ceased firing abruptly and in these cases the impulse frequency–pressure relationship remained linear.

In multi-fibre preparations of the aortic nerve it was found that as the aortic arch pressure was raised recruitment occurred of other fibres which had higher threshold pressures.

Right aortic nerve

The results of a study of ten fibres from the right aortic nerve in five experiments are summarized in Table 1 and the relationship between the impulse frequency and intra-aortic pressure is illustrated in Fig. 3 (right).

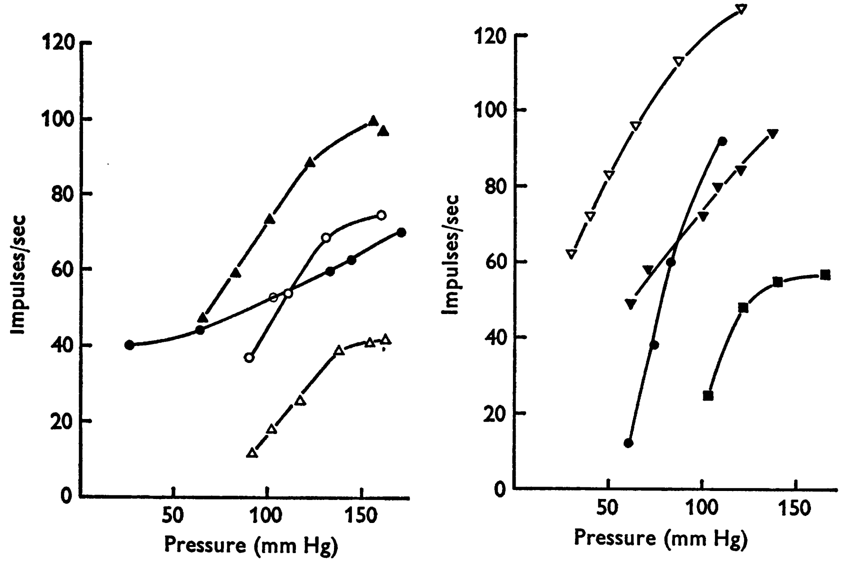

Fig. 3. Graphs showing the relationship of impulse activity to intra-aortic pressure. *Left*, four single fibres from the left aortic nerve. *Right*, four single fibres from the right aortic nerve. The lowest point of each curve represents the threshold pressure of each receptor.

The nature of the firing at threshold levels of pressure was classified according to the three types described above and seven fibres were of Type 1, one of Type 2 and two of Type 3.

Comparison of left and right aortic nerves. Table 1 also shows that for the two nerves there is little difference between the corresponding values for the threshold index, the slope of the initial part of the impulse frequency–pressure curve, the point of inflexion, the impulse frequency at the point of inflexion, and the index at the point of inflexion. In subsequent sections of this paper, therefore, the characteristics of the fibres comprising these two nerves will be considered together.

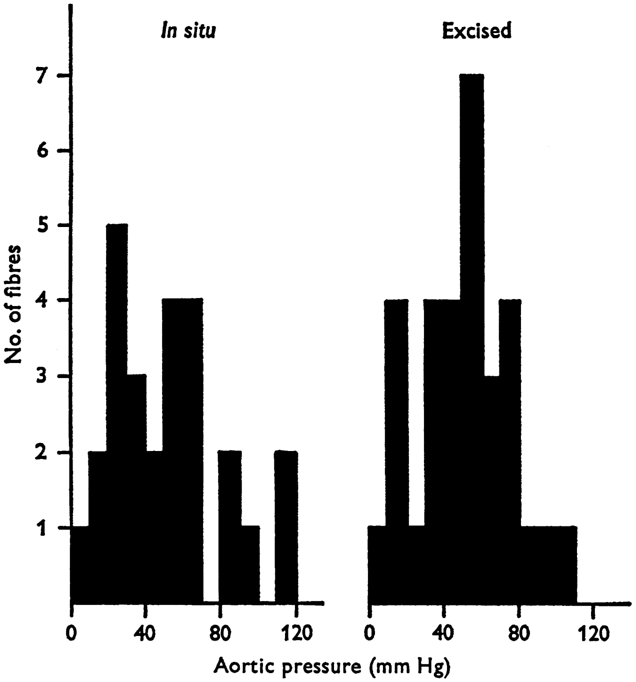

Fig. 4. Histograms showing the threshold pressure (mm Hg) of twenty-six fibres from the left aortic nerve with the aortic arch preparation *in situ* (left) and thirty-one fibres with the excised aortic arch preparation (right).

Effects of altering the composition and temperature of the perfusate

Composition. In the majority of experiments Krebs–Henseleit solution was used as the perfusate and it was therefore considered necessary to find out whether the impulse frequency–pressure curves differed in any way when blood was used. The effects of substituting dog's blood for Krebs–Henseleit solution (eight fibres in four experiments) and rabbit's blood for Krebs–Henseleit solution (two fibres in two experiments) were therefore studied. No appreciable difference between any of the curves was found, and the results from a typical experiment are shown in Fig. 5.

Temperature. The effects of lowering the temperature of the perfusate on the relationship between impulse frequency and aortic arch pressure was studied in six experiments at three different temperatures (mean values 38·2, 29·0 and 22·1° C). In order for the comparison to be made the impulse frequency was determined at the threshold pressure and at pressures of 60, 80 and 100 mm Hg. The results obtained from thirteen fibres, of which five were from the right aortic nerve and eight from the left, are shown graphically in Fig. 6, from which it may be seen that as the temperature is lowered the impulse frequency at a given aortic pressure is reduced.

A paired analysis of the data was made by comparing the values at the two lower temperatures with the corresponding control values at 38·2° C. Compared with the control temperature the threshold pressure and the impulse frequency at the threshold pressure were respectively 11·5 ± 4·1 mm Hg and 6·5 ± 2·9 impulses/sec lower at the temperature of 29·0° C ($P < 0{\cdot}02$ and $P < 0{\cdot}05$) and 13·9 ± 5·0 mm Hg and 10·7 ± 2·6 impulses/sec lower at the temperature of 22·1° C ($P < 0{\cdot}02$ and $P < 0{\cdot}001$). The reduction in impulse frequency at pressures of 60, 80 and 100 mm Hg is also statistically significant ($P < 0{\cdot}001$) and becomes greater as the pressure increases (see Fig. 6).

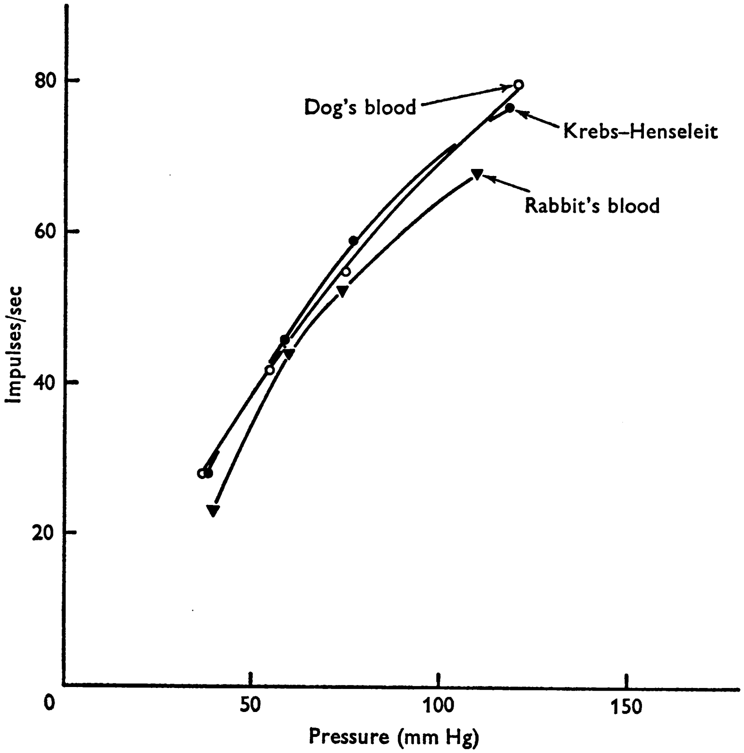

Fig. 5. Graph relating the impulse activity from a single fibre of the right aortic nerve to aortic arch pressure. The temperature of the perfusates was the same. ●, Krebs–Henseleit solution; ○, dog's blood; ▼, rabbit's blood.

When the temperature of the perfusate was lowered the gradient of the first part of the curve relating impulse frequency to intra-aortic pressure was depressed from a mean of 1·56 ± 0·29 impulses/sec.mm Hg (range 0·36–3·76) at 38·2° C to a mean of 0·74 ± 0·16 impulses/sec.mm Hg (range 0·18–1·75) at 29·0° C and 0·22 ± 0·1 impulses/sec.mm Hg (range − 0·28 to + 1·0) at 22·1° C. In some experiments it was found that with the lowest temperature of 22·1° C the slope was negative, indicating a fall of the impulse frequency as the intra-aortic pressure increased. Some fibres even became inactive at a perfusion temperature of 22° C.

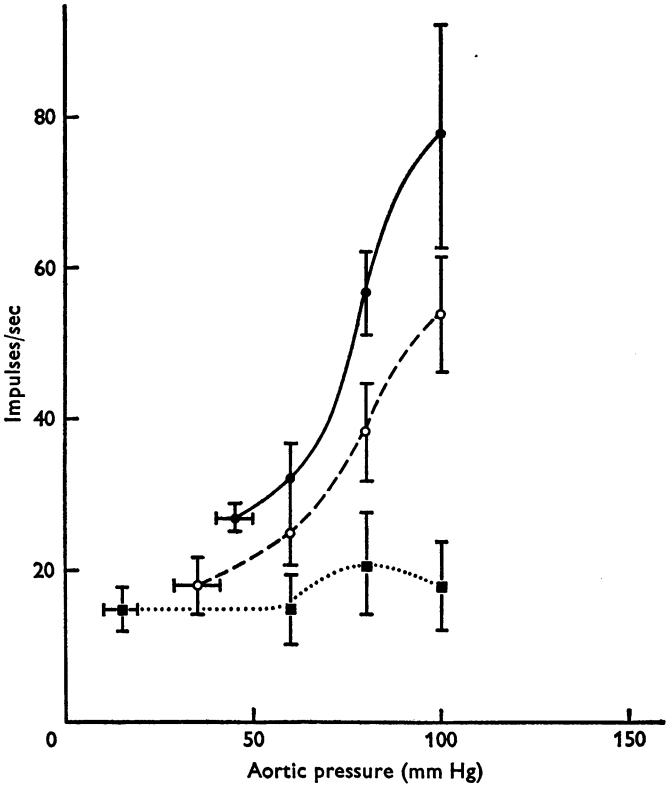

Fig. 6. The effect of temperature on the relationship of impulse activity to aortic arch pressure. Thirteen fibres of the aortic nerves. Perfusate: Krebs–Henseleit solution. ●—●, temperature 38·2° C; ○---○, temperature 29·0° C; ■....■, temperature 22·1° C. Values are the means ± S.E. of mean.

The results obtained on another four fibres (two from the left and two from the right aortic nerves) in two experiments are not included in Fig. 6 as their curves relating impulse frequency to intra-aortic pressure lay outside the pressure range used for this particular analysis. However, they exhibited a similar reduction of impulse frequency at any given pressure when the temperature of the perfusate was lowered.

The effect on aortic baroreceptor activity of reducing aortic pressure in steps after previously raising the pressure

A comparison was made of the relationship between the impulse frequency and aortic arch pressure during a stepwise increase in aortic pressure and during similar decrease in pressure. It was found in all six experiments that the two curves differed slightly in position (Table 2). The typical effect is shown in Fig. 7. It may be observed that whereas the curves at the higher levels of pressure did not differ appreciably, the lower part of the descending pressure curve fell to the right of the ascending pressure curve. The lowest pressure at which impulses were recorded on the descending pressure curve was 53 mm Hg, compared with the original 'threshold' pressure of 41 mm Hg on the ascending pressure curve. The frequency of firing was also less than for the equivalent pressure when the pressure was progressively raised, and the threshold index was therefore reduced.

TABLE 2. A comparison of the relationship between the impulse frequency and aortic arch pressure during (*A*) a stepwise increase and (*B*) a stepwise decrease in aortic arch pressure. Six fibres in six experiments

	A (increasing pressure)	*B* (decreasing pressure)	Difference (*B* − *A*)
Threshold pressure (mm Hg)	56·6 ± 4·8 (20–104)	69·2 ± 11·7 (33–104)	12·5 ± 8·9 (−10 to +50) $P > 0·2$
Threshold frequency (impulses/sec)	32·3 ± 5·9 (8–50)	30·0 ± 8·2 (1–60)	−2·3 ± 3·3 (−11 to +10) $P > 0·5$
Threshold index (impulses/sec. mm Hg)	0·98 ± 0·38 (0·08–2·5)	0·54 ± 0·16 (0·01–1·03)	−0·62 ± 0·32 (−1·64 to +0·04) $0·1 > P > 0·05$
Frequency at 20 mm Hg above threshold pressure (impulses/sec)	48·2 ± 5·4 (27–63)	33·2 ± 5·0 (19–50)	−12·4 ± 2·4 (−18 to −6) $P < 0·01$
Frequency at 40 mm Hg above threshold pressure (impulses/sec)	57·0 ± 7·0 (34–77)	46·4 ± 6·1 (27–62)	−7·0 ± 2·6 (−15 to 0) $P < 0·02$
Frequency at 60 mm Hg above threshold pressure (impulses/sec)	60·2 ± 7·9 (45–90)	56·0 ± 4·7 (47–74)	−4·2 ± 3·2 (−16 to +2) $P > 0·2$
Gradient (impulses/ sec. mm Hg)	1·07 ± 0·4 (0·28–2·77)	1·37 ± 0·36 (0·36–2·77)	0·30 ± 0·42 (−0·04 to +2·17) $P > 0·5$

The open values are the means ± S.E. of mean. The ranges are in parentheses

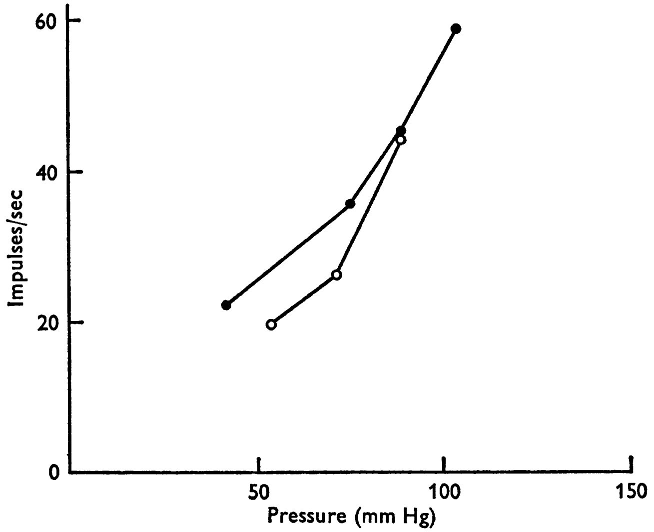

Fig. 7. Graphs relating the impulse activity of a single fibre from the left aortic nerve to aortic pressure. The aortic arch of a rabbit, wt. 1·5 kg, was isolated and perfused *in situ* with Krebs–Henseleit solution. ●, Increasing the pressure; (○), decreasing the pressure.

II. The excised aortic arch preparation

As certain investigations reported elsewhere (Angell James, 1971*b*) could only be carried out on the excised aortic arch preparation, it was necessary to compare the characteristics of the baroreceptors in this preparation with those observed in the *in situ* preparation.

Left aortic nerve

The relationship between impulse frequency and aortic arch pressure was studied in thirty-one single- or few-fibre excised preparations and the results are summarized in Table 1. The threshold pressures, also depicted in the histogram in Fig. 4, ranged from 15 to 110 mm Hg (mean 52·9 ± 4·5). The nature of the firing at threshold pressure was variable, twenty-four fibres being of Type 1, six of Type 2 and one of Type 3. Over the range of pressures used, only fifteen of the thirty-one curves reached a point of inflexion. The remaining sixteen (51·6 %) fibres continued to maintain a linear relationship between impulse frequency and aortic pressure.

Comparison with in situ *preparations.* A comparison was made of this data (Table 1) with that obtained from the examination of twenty-six fibres in fifteen experiments carried out on the *in situ* preparations. Group analysis of the data indicates that there are no differences in threshold pressure ($P > 0·7$) or the impulse frequency at the threshold pressure

($P > 0.2$). The threshold index, however, was lower. There was a significant difference between the values for the gradient of the first part of the curves, 1·11 ± 0·14 impulses/sec.mm Hg in the *in situ* preparations and 1·58 ± 0·05 impulses/sec.mm Hg in the excised preparations ($P < 0.05$).

The data for comparison of the points of inflexion in the two series is more difficult to evaluate. Although the mean pressure corresponding to the point of inflexion was less in the 'excised' preparations the data represented only 48·4 % of the fibres, as the remainder failed to reach this point. This compares with 61·5 % of the fibres in the *in situ* preparations. Thus a smaller proportion of fibres reached their point of inflexion in the 'excised' preparation and those fibres that did had a higher frequency of impulses than those in *in situ* preparations. The index at the point of inflexion was found to be less in the *in situ* preparations than in the 'excised' preparations. This indicates that the frequency of impulses was higher at the point of inflexion in the 'excised' preparation.

Right aortic nerve

In the 'excised' preparations of the aortic arch and subclavian areas the threshold pressure was similar to that in the *in situ* preparations, but of ten fibres studied only two showed a point of inflexion. The remaining eight fibres (80 %) did not reach this point.

Comparison with in situ *preparations*. Group comparison of the data for the right aortic nerve obtained from *in situ* and 'excised' aortic arch preparations indicates that there is no difference between the values for threshold pressure ($P > 0.8$), impulse frequency at the threshold pressure ($P > 0.1$) or gradient of the first part of the curve ($P > 0.5$) (Table 1). The threshold index, however, was lower in the excised preparation.

No detailed comparison could be made of the values for the point of inflexion in the two series as only two of the ten fibres (20 %) in the 'excised' aortic arch reached this point; in the *in situ* preparation 60 % of the fibres showed this point at a mean pressure of 97·7 ± 5·5 mm Hg and mean impulse frequency of 65·5 ± 5·3 impulses/sec. However, the fact that 80 % of the fibres in the 'excised' preparation compared with 40 % in the *in situ* preparation failed to reach their point of inflexion in the range of pressures used indicated that the characteristics of the baroreceptors in the 'excised' preparation are different to those in the *in situ* preparation.

It is apparent that the differences in the relationship between impulse frequency and aortic arch pressure in the *in situ* and 'excised' preparation, particularly with regard to the position of the point of inflexion, apply to fibres of both the left and right aortic nerves.

The different behaviour of the baroreceptors in the two types of preparation may be due to the fact that in the 'excised' preparation cutting the vessels released the longitudinal elastic tension of the aortic arch and common carotid–subclavian angle. However the aortic arch is also free to straighten in this preparation, which would alter the radii of curvature and modify the general form of the Laplace equation. In order to study the effect of releasing the longitudinal tension alone on single fibre activity, further experiments were carried out as described below.

III. The effects of relasing the longitudinal tension in the arterial walls

Experiments were carried out on the aortic arch *in situ* and impulse frequency–aortic arch pressure curves were obtained before and after releasing the longitudinal tension of the aortic arch. This was done by cutting transversely the descending aorta distal to the ligature on the vessel (see Methods). In this series visual inspection indicated there was little alteration of the curvature of the aortic arch, presumably due to the tethering to adjacent tissues being undisturbed.

A typical experiment is illustrated in Fig. 8, in which the impulse discharge in a single fibre of the left aortic nerve was studied. After cutting the descending aorta the threshold pressure was elevated but the impulse frequency at the threshold pressure did not increase correspondingly; on the contrary, it fell below the control level. Thus the threshold index was reduced from 1·5 to 0·43 impulses/sec.mm Hg. The point of inflexion was increased by 40 mm Hg after severing the aorta and the impulse frequency was augmented at these pressures.

The results of four experiments in which five fibres were studied before and after severing the descending aorta (Table 3) showed that this procedure resulted in an elevation of the threshold pressure in four of the five fibres; but the fifth fibre now exhibited a Type 3 threshold so that a constant rate of firing was maintained over a range of pressure from 17 mm Hg below the control threshold to 15 mm Hg above it. The average value for the threshold was raised, but there was no increase in either the impulse frequency, despite this elevation of the threshold pressure, or the threshold index. Reducing the longitudinal tension on the vessels also caused an increase in the pressure at which the point of inflexion occurred and an increase in the impulse frequency at the point of inflexion. However, there was no difference between the mean control and experimental values for the gradient of the linear part of the curve.

It is concluded from these experiments that severing the aortic vessels alters the relationship between the impulse frequency and aortic pressure. This corroborates the findings that the relationship is also different in ‘excised’ and *in situ* aortic-arch preparations.

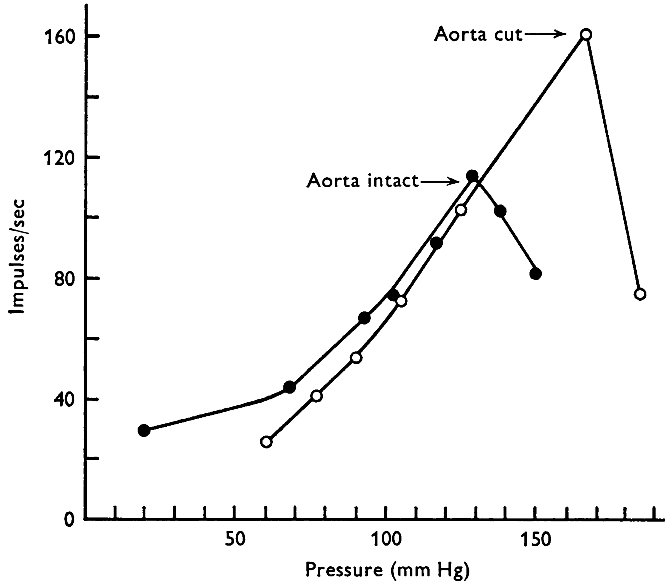

Fig. 8. Graphs showing the relationship between impulse activity and aortic pressure. Single fibre from the left aortic nerve of a 1·6 kg rabbit. The aortic arch was isolated and perfused with Krebs-Henseleit solution. ●, aortic arch *in situ*; ○, aortic arch *in situ* and the aorta divided distal to the origin of the left subclavian artery.

Table 3. The effect on the behaviour of the aortic arch baroreceptors of altering the longitudinal tension of the aortic arch by cutting the vessel. Five fibres in four experiments. Aortic arch preparation *in situ*

	Control (*A*)	Experimental (*B*)	Difference (*B* − *A*)
Threshold pressure (mm Hg)	40·2 ± 12·8 (13–84)	47·8 ± 13·8 (15–88)	+ 7·2 ± 9·1 (− 17 to + 40) $P > 0·4$
Threshold frequency (impulses/sec)	33·6 ± 1·7 (30–39)	31·8 ± 2·8 (24–39)	− 2·0 ± 1·8 (− 6 to + 4) $P > 0·3$
Threshold index (impulses/sec.mm Hg)	1·2 ± 0·33 (0·41–2·31)	1·01 ± 0·36 (0·43–2·3)	− 0·2 ± 0·37 (− 1·07 to + 1·2) $P > 0·6$
Point of inflexion (mm Hg)	98·2 ± 10·8 (63–129)	138·6 ± 11·5 (113–167)	30·7– ± 3·7 (27–38) $P < 0·01$
Frequency at point of inflexion (impulses/sec) (three fibres)	81·4 ± 11·1 (51–115)	133 ± 14·5 (87–160)	36·3 ± 9·7 (17–47) $P < 0·02$
Gradient (impulses/sec.mm Hg)	1·4 ± 0·37 (0·86–2·85)	1·3 ± 0·25 (0·86–2·2)	− 0·09 ± 0·16 (− 0·65 to + 0·34) $P > 0·5$

Two fibres failed to reach the point of inflexion after severing the aortic arch. The open values are the means ± s.e. of mean. The ranges are in parentheses.

DISCUSSION

The isolated aortic-arch preparation described here has enabled a study to be made of the physiological characteristics of the baroreceptors in the aortic-arch and right subclavian areas under conditions of controlled perfusion. When perfused with non-pulsatile pressures no systematic differences were found as indicated by the characteristics of the impulse frequency–aortic arch pressure curves. This is consistent with the observations of Irisawa & Ninomiya (1967), who found no difference in the 'form' of the whole nerve activity in the left and right aortic nerves of the rabbit.

The threshold pressure of the aortic arch baroreceptors lay in the range given by experiments *in vivo* in which thresholds were calculated by the use of regression lines from data relating impulse activity to mean arterial blood pressure in the anaesthetized rabbit (Bloor, 1964). Bauer (1939) was unable to obtain any reflex effect from stimulation of these receptors until a pressure of 60–65 mm Hg was obtained. However, any comparison between his values and those reported here is difficult to assess because the data from the present experiments is based on receptor threshold whereas Bauer (1939) studied the threshold for the reflex response which would include the characteristics of the central and peripheral parts of the autonomic nervous system.

The threshold pressure for the right subclavian baroreceptors was the same as for the receptors in the aortic arch. Other workers have studied the threshold for arterial blood pressure response from the right subclavian baroreceptor areas perfused at non-pulsatile pressures, and whereas Tang (1964) obtained values of 60–80 mm Hg, those observed by Ueda, Uchida, Yasuda & Takeda (1966) were lower (20 ± 8·6 mm Hg).

The phenomenon of impulse activity occurring sometimes at very low pressures was also observed in the carotid sinus by Landgren (1952*a*), who ascribed it to deformation of the receptor endings by collapse of the arterial wall. A similar explanation for the reflex effect observed in the low-pressure ranges was given by Heymans & Bouckaert (1929) to account for the fall in blood pressure which occurred on lowering the carotid sinus pressure from an initial level of about 50 mm Hg. However, in the present experiments it appears that some fibres fire constantly over a pressure range extending to 0 mm Hg and this must be due to a constant tension acting on the nerve endings. Occasionally a slight increase in discharge occurred as the pressure approached 0 mm Hg, which could be due to the collapse of the arterial wall deforming the receptors.

The explanation for the three types of threshold described in this paper must lie in the nature of the intimate connexions of the receptors with the smooth muscle, elastic and collagen tissues in the wall; thus the intermittent (Type 2) threshold could be due to phasic changes in tone of the vessel wall and the constant discharge (Type 3) threshold to a constant tension acting on the receptor endings over that pressure range.

The linear relationship between the arterial pressure and impulse frequency in the lower parts of the curves relating these two parameters was described for the carotid sinus baroreceptors by Bronk & Stella (1932, 1935). This is the most sensitive part of the curve and straddles the value for the normal mean arterial blood pressure for the unanaesthetized rabbit (92 ± 5·8 mm Hg, Chalmers, Korner & White, 1967). The curves obtained by Homma & Suzuki (1966) in the cat were not only less steep than those reported here but were also S-shaped, thus differing from the majority of fibres in the present series. The possibility that the aortic arch was 'splinted' by the clamps applied to the proximal and distal parts of the arch in their experiments might account for this difference, particularly in view of the observed changes in the shape of the curves that occur on deliberately altering the longitudinal tension in the aorta (this paper).

The impulse frequency at the point of inflexion was not necessarily the maximum frequency attained in any given fibre; in some fibres, at least, the impulse frequency continued to increase with pressure whereas in others the impulse frequency either remained constant, diminished, became intermittent or decreased to zero. The enormous variation in the response between one receptor and another may be due to their position in the wall of the vessel and to their relationship to different tissues within the wall.

The shape of the impulse frequency–aortic pressure curve was also dependent on the previous history of the preparation. Thus following distension of the aortic arch the lower part of the curve is shifted to the right, resulting in a diminished frequency of impulse activity at a given pressure. This indicates an alteration of receptor activity most probably due to mechanical changes resulting from the arterial wall having been stretched (static hysteresis).

Studies based on the aortic-arch baroreceptor impulse activity and on their efferent response indicate that the most sensitive control of fluctuations of arterial blood pressure would occur at levels of pressure found in the normal animal (Angell James & Daly, 1970). Where the arterial pressure gradually falls due, for instance, to haemorrhage, the impulse frequency in individual fibres diminishes and there is, in addition, a progressive reduction in the number of active units. As the pressure rises above normal, the incremental increase in the total number of impulses becomes progressively less, due to the point of inflexion being reached in individual units when the curve begins to flatten and, in the case of some receptors at least, starts to fall. Few fibres would be recruited. Whole nerve recordings of the aortic nerves in the intact animal confirm that at pressures above about 140 mm Hg in the rabbit there is little increase in total nerve activity (Aars, 1968).

Effects of hypothermia

The reversible diminution in the frequency of impulses in single fibres of both the left and right aortic nerves at any given pressure produced by lowering the temperature of the perfusate is an observation similar to that made by Diamond (1955) on the carotid sinus of the cat. The only difference appears to be in connexion with the effect of temperature on the threshold pressure. Whereas the threshold, at least in the case of some of the aortic fibres, was reduced by lowering the temperature of the perfusate, Diamond (1955) found it to be increased in carotid sinus fibres.

The possibility must be considered that the effects of hypothermia on the aortic baroreceptor impulse activity is the result of partial block of nerve transmission. However, the evidence presented by Paintal (1965*a*, *b*) indicates that even at the minimum temperature used, 26·5° C, and at impulse frequencies of the order of 100 impulses/sec, this explanation is unlikely. Alternative explanations are that the phenomenon is due to depolarization of the membrane by the nerve endings, such as occurs with muscle spindles (Lippold, Nicholls & Redfearn, 1960), or to an alteration in the visco-elastic properties of the wall of the blood vessel. With regard to the last possibility, cooling blood vessels results in contraction of smooth muscle and in a reduction in distensibility. At any given pressure, therefore, the radius of the aortic arch is less on cooling than at a temperature of 37° C. No experiments have been carried out to determine which of the two alternatives is the more likely.

These results have a bearing on the arterial baroreceptor reflex responses under conditions of hypothermia. Nashat & Neil (1955) found that reducing the temperature of the carotid sinus perfusate, the body temperature remaining normal, reduced the arterial blood pressure response to a given rise of carotid sinus pressure.

Possible mechanisms of stimulation of arterial baroreceptors

Hauss, Kreuziger & Asteroth (1949) demonstrated that the reflex fall in blood pressure resulting from an increase in carotid sinus pressure was dependent on stretch, 'distension' or deformation of the sinus wall. Consistent with this view is the finding that the increased aortic baroreceptor impulse activity resulting from a rise in aortic arch pressure can be prevented by simultaneously applying an extra-mural counter-pressure of similar size (Angell James, 1971*b*). Moreover the shape of the curve relating impulse activity to aortic arch pressure resembles the pressure–volume curves (Hallock & Benson, 1937; Remington, 1955), the point of inflexion occurring at a point when the volume or circumference ceases to increase linearly with pressure. Excision of the preparation results in an elevation of the point of inflexion and the frequency of impulses which is probably due to the release of the longitudinal tension allowing an additional increase in volume, which has been shown to occur, at higher pressures when the aorta is excised (Bergel, 1961). This suggests there is a close relationship between aortic volume and baroreceptor activity. This is supported by the work of Aars (1969) in which he demonstrated a close relationship between baroreceptor activity, from whole nerve recordings, and aortic diameter.

It is well known that the pressure–circumference relationship is dependent on the longitudinal tension, a reduction in longitudinal tension resulting in an increased circumference at a given pressure (Rushmer, 1955; Bergel, 1961). It might be expected, therefore, that if the baroreceptor fibre impulse frequency was solely a function of vessel diameter, the physiological effects of dividing the aortic arch would be to increase the impulse frequency at a given pressure. But this was not always so, except at a pressure above that for the normal inflexion point. This finding can be reconciled if it is assumed that the attachments of the receptors in relation to the components of the vessel wall are such that the receptors are stretched not only by an increase in circumference of the vessel, but also by an increase in its longitudinal length. In this connexion traction on the common carotid artery stimulates the sinus nerve endings (Heymans & Neil, 1958, p. 76). The explanation for the elevation of the threshold pressure which occurs after division of the aorta may be similar. Thus after reducing the longitudinal tension on the aortic arch, the impulse frequency at a given pressure would be the resultant of an increased rate of firing through an increase in circumference and a reduced discharge due to a diminished longitudinal length.

One of the striking features of the present study was the variability of the characteristics of the aortic-arch baroreceptors as found by Bronk & Stella (1935) for the carotid sinus baroreceptors. This may be a reflexion of the wide range of contact which the nerve terminals make with individual components of the arterial wall and various admixtures of these components in a particular area, as suggested for the carotid sinus by Rees (1967). No relationship was found between action potential height and specific characteristics of the curves. In any case action potential height depends not only on aortic nerve fibre diameter (2–10 μ, Paintal, 1953) but also on the distance of the active fibre from the recording electrode.

There is evidence that in addition to changes of vessel circumference and longitudinal stretch, the baroreceptors are also affected by local mechanical changes within the vessel wall. The local application of adrenaline or noradrenaline to the carotid sinus causes a reflex fall in arterial blood pressure (Palme, 1951; Heymans & Heuvel-Heymans, 1951). Heymans & Heuvel-Heymans postulated that this response was due to an increase in tone or tension within the vessel wall which affected baroreceptors. Landgren (1952*b*) demonstrated the increase baroreceptor activity by electroneurographic recordings and found that it was accompanied by a *reduction* in diameter of the carotid sinus. Further evidence that local mechanical changes in the vessel affect baroreceptors is presented in the present paper.

In the construction of static impulse–frequency–pressure curves, as for the dynamic curves (Angell James, 1971*a*), the impulse frequency was found to be lower at a given pressure on the descending pressure limb despite the aortic diameter being larger (Remington, 1955). The explanation of these effects is not entirely clear but certain aspects of the problem have been discussed previously (Peterson, 1960; Bader, 1963).

On the basis of the views of Benninghoff (1930) on the structural relationship between smooth 'spannung' muscles and elastic membranes and fibres in the wall of elastic vessels, contraction of the smooth muscle, for instance by adrenaline, would increase the tension, and hence the length of the elastic fibres and so stimulate the baroreceptors lying parallel with these fibres (see Bader, 1963). Such an explanation would account for the changes occurring in the static impulse frequency–pressure loops if changes occurred predominantly in the smooth muscle component of the vessel wall. Alternatively it is known that the relative extensibilities of the various components of the vessel wall differ considerably (Burton, 1954). Thus a change in extensibility in one component relative to another at the same pressure may alter the activity in adjacent baroreceptors as some of the nerve terminals situated near the external elastic lamina are related to collagen and elastic tissue, at least in the carotid sinus (Rees, 1967). In the aortic arch receptors are also found in the outer third of the medial coat (Köster & Tschermak, 1902; Nonidez, 1935) and are therefore affected by the mechanical properties of this area of the vessel wall in addition to those of the external elastic limina.

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